Genetic Overlap Between Attention-Deficit/Hyperactivity Disorder and Bipolar Disorder: Evidence From Genome-wide Association Study Meta-analysis

van Hulzen, Kimm J E ; Scholz, Claus J ; Franke, Barbara ; Ripke, Stephan ; Klein, Marieke ; McQuillin, Andrew ; Sonuga-Barke, Edmund J S ; PGC ADHD Working Group ; Kelsoe, John R ; Landén, Mikael ; Andreassen, Ole A ; et al ; Walitza, Susanne ; Steinhausen, Hans-Christoph ; PGC Bipolar Disorder Working Group ; Lesch, Klaus-Peter ; Weber, Heike ; Faraone, Stephen V ; Arias-Vasquez, Alejandro ; Reif, Andreas

Abstract: Background Attention-deficit/hyperactivity disorder (ADHD) and bipolar disorder (BPD) are frequently co-occurring and highly heritable mental health conditions. We hypothesized that BPD cases with an early age of onset (21 years old) would be particularly likely to show genetic covariation with ADHD. Methods Genome-wide association study data were available for 4609 individuals with ADHD, 9650 individuals with BPD (5167 thereof with early-onset BPD), and 21,363 typically developing controls. We conducted a cross-disorder genome-wide association study meta-analysis to identify whether the observed comorbidity between ADHD and BPD could be due to shared genetic risks. Results We found a significant single nucleotide polymorphism–based genetic correlation between ADHD and BPD in the full and age-restricted samples (rGfull = .64, p = 3.13 × 10–14; rGrestricted = .71, p = 4.09 × 10–16). The meta-analysis between the full BPD sample identified two genome-wide significant (prs7089973 = 2.47 × 10–8; prs11756438 = 4.36 × 10–8) regions located on chromosomes 6 (CEP85L) and 10 (TAF9BP2). Restricting the analyses to BPD cases with an early onset yielded one genome-wide significant association (prs58502974 = 2.11 × 10–8) on chromosome 5 in the ADCY2 gene. Additional nominally significant regions identified contained known expression quantitative trait loci with putative functional consequences for NT5DC1, NT5DC2, and CACNB3 expression, whereas functional predictions implicated ABLIM1 as an allele-specific expressed gene in neuronal tissue. Conclusions The single nucleotide polymorphism–based genetic correlation between ADHD and BPD is substantial, significant, and consistent with the existence of genetic overlap between ADHD and BPD, with potential differential genetic mechanisms involved in early and later BPD onset.

DOI: https://doi.org/10.1016/j.biopsych.2016.08.040

Originally published at:
van Hulzen, Kimm J E; Scholz, Claus J; Franke, Barbara; Ripke, Stephan; Klein, Marieke; McQuillin, Andrew; Sonuga-Barke, Edmund J S; PGC ADHD Working Group; Kelsoe, John R; Landén, Mikael; Andreassen, Ole A; et al; Walitza, Susanne; Steinhausen, Hans-Christoph; PGC Bipolar Disorder Working Group; Lesch, Klaus-Peter; Weber, Heike; Faraone, Stephen V; Arias-Vasquez, Alejandro; Reif, Andreas (2017). Genetic Overlap Between Attention-Deficit/Hyperactivity Disorder and Bipolar Disorder: Evidence From Genome-wide Association Study Meta-analysis. Biological Psychiatry, 82(9):634-641.
DOI: https://doi.org/10.1016/j.biopsych.2016.08.040

Biological Psychiatry

Archival Report

Genetic Overlap Between Attention-Deficit/Hyperactivity Disorder and Bipolar Disorder: Evidence From Genome-wide Association Study Meta-analysis

Kimm J.E. van Hulzen, Claus J. Scholz, Barbara Franke, Stephan Ripke, Marieke Klein, Andrew McQuillin, Edmund J. Sonuga-Barke, PGC ADHD Working Group, John R. Kelsoe, Mikael Landén, Ole A. Andreassen, PGC Bipolar Disorder Working Group, Klaus-Peter Lesch, Heike Weber, Stephen V. Faraone, Alejandro Arias-Vasquez, and Andreas Reif

ABSTRACT

BACKGROUND: Attention-deficit/hyperactivity disorder (ADHD) and bipolar disorder (BPD) are frequently co-occurring and highly heritable mental health conditions. We hypothesized that BPD cases with an early age of onset (≤21 years old) would be particularly likely to show genetic covariation with ADHD.

METHODS: Genome-wide association study data were available for 4609 individuals with ADHD, 9650 individuals with BPD (5167 thereof with early-onset BPD), and 21,363 typically developing controls. We conducted a cross-disorder genome-wide association study meta-analysis to identify whether the observed comorbidity between ADHD and BPD could be due to shared genetic risks.

RESULTS: We found a significant single nucleotide polymorphism–based genetic correlation between ADHD and BPD in the full and age-restricted samples ($r_{Gfull} = .64$, $p = 3.13 \times 10^{-14}$; $r_{Grestricted} = .71$, $p = 4.09 \times 10^{-16}$). The meta-analysis between the full BPD sample identified two genome-wide significant ($p_{rs7089973} = 2.47 \times 10^{-8}$; $p_{rs11756438} = 4.36 \times 10^{-8}$) regions located on chromosomes 6 (*CEP85L*) and 10 (*TAF9BP2*). Restricting the analyses to BPD cases with an early onset yielded one genome-wide significant association ($p_{rs58502974} = 2.11 \times 10^{-8}$) on chromosome 5 in the *ADCY2* gene. Additional nominally significant regions identified contained known expression quantitative trait loci with putative functional consequences for *NT5DC1*, *NT5DC2*, and *CACNB3* expression, whereas functional predictions implicated *ABLIM1* as an allele-specific expressed gene in neuronal tissue.

CONCLUSIONS: The single nucleotide polymorphism–based genetic correlation between ADHD and BPD is substantial, significant, and consistent with the existence of genetic overlap between ADHD and BPD, with potential differential genetic mechanisms involved in early and later BPD onset.

Keywords: Attention-deficit/hyperactivity disorder, bipolar disorder, cross-disorder meta-analysis, genetic correlation, genetic overlap, GWAS

http://dx.doi.org/10.1016/j.biopsych.2016.08.040

Attention-deficit/hyperactivity disorder (ADHD) is the most frequent neuropsychiatric disorder in childhood and frequently persists into adulthood. Bipolar disorder (BPD) is among the most prevalent mental diseases in adulthood. Both disorders are highly heritable (1,2). However, in both cases the mode of inheritance is complex and polygenic (3). Although differing from one another with regard to core signs and symptoms, age of onset, presentation, and treatment response, the two disorders share several clinical features. This is especially the case for the manic phase of BPD, which is associated with irritability, increased impulsivity, distractibility, and restlessness (4). Furthermore, ADHD often copresents with depression (5), a core feature of BPD. In adulthood, when BPD is most commonly diagnosed, co-occurrence of the two disorders occurs more often than would be expected by chance (6). For patients with BPD rates of ADHD vary between 9.5% and 28%, depending on study characteristics (7,8). The rate of BPD in adult ADHD has been estimated at around 20% (7). Meta-analyses of family studies confirm elevated rates of BPD in first-degree relatives of ADHD patients and vice versa (8).

Although a shared genetic basis for ADHD and BPD seems plausible given the above, molecular genetic studies thus far provide limited evidence for this (9–11). For instance, risk-allele frequencies of candidate genes, identified through prior ADHD genome-wide association studies (GWASs), are not increased in BPD. This failure to find a shared genetic signal may be due to prior studies' lack of statistical power and to the limited set of polymorphisms examined. To address these shortcomings,

SEE COMMENTARY ON PAGE 621

 ISSN: 0006-3223

a genome-wide cross-disorder meta-analysis of BPD and ADHD was conducted in a large sample of individuals with BPD, indivduals with ADHD, and in typically developing control subjects from the Psychiatric Genomics Consortium. Because ADHD is a childhood onset disorder (prior to 12 years of age), we hypothesized that the overlap would be most obvious in BPD cases with a relatively early onset (age of onset ≤21 years old), as this group could be assumed to have a more neurodevelopmental etiology (8). Restricting the age range of the sample to those with an onset ≤21 years of age may also increase the power for gene finding because it could reduce heterogeneity (12). We thus performed analyses with the total number of BPD cases as well as the age-restricted set. GWAS meta-analysis top findings were further characterized using expression quantitative trait locus (eQTL) analysis to investigate potential functional consequences for gene expression.

METHODS AND MATERIALS

Samples

Cases, controls, and family-based samples assembled for previous genome-wide Psychiatric Genomics Consortium analyses of individual-level data were included in the current analysis (13,14). A description of individual study data contributions and genotyping platforms is included in Supplemental Tables S1A and S1B. The ADHD sample comprised 4609 cases and 8519 controls. The full BPD sample comprised 9650 cases and 12,844 controls. For tests of the age of onset hypothesis we restricted the BPD sample to cases with an age of onset ≤21 years of age. This reduced the number of cases to 5167 (restricted sample). All available controls from the BPD samples were included in the age-restricted sample to maximize power. Control individuals that featured in both the ADHD and BPD samples were identified and removed prior to analysis.

Genetic Analysis

Raw genotype and phenotype data for each study was uploaded to a central server and processed through the same quality control, imputation, and analysis process to ensure comparability between the samples. The quality control and analysis pipeline is described elsewhere (3).

Statistical Analysis

Linkage disequilibrium (LD) score regression was used to estimate the single nucleotide polymorphism (SNP)–based genetic correlation (r_G) between the ADHD and both BPD samples. For LD score regression, each data set underwent additional filtering. Only markers overlapping with HapMap Project Phase 3 SNPs and passing the following filters were included: INFO (imputation) score > 0.9, study missingness of 0, and minor allele frequency >1% (where available). Indels and strand-ambiguous SNPs were removed.

The analysis was conducted using a two-step procedure with the LD scoring analysis package (https://github.com/bulik/ldsc) (15). An unconstrained regression was run to estimate the regression intercepts for each phenotype, followed by an analysis with regression intercepts constrained to those estimated in the first step and an unconstrained covariance intercept (we took steps to exclude overlapping samples). Standard errors were estimated using a block jackknife procedure and used to calculate p values.

GWAS was initially performed for each ADHD study separately (n = 8). Four multidimensional scaling components were included to account for potential population stratification. GWAS was then also performed for each BPD study separately (n = 12). In this case a total of seven multidimensional scaling components (both total and restricted samples) were used in the analysis in order to correct for potential population stratification. These GWASs were free from genomic inflation as judged by quantile-quantile plots (data not shown). For each disorder, results were then combined in a disorder-specific meta-analysis. Finally, results from the disorder-specific meta-analyses were combined in cross-disorder meta-analyses for both the primary and the age-restricted samples. For all meta-analyses, we applied a weighted Z-score approach using PLINK 1.07 (http://pngu.mgh.harvard.edu/~purcell/plink/index.shtml), in which weights equaled the inverse of the regression coefficient's standard error (16). This strategy assumed a fixed-effects model, in which all studies/disorders had the same direction of effect, with weights indicating the sample size and imputation accuracy of the disease-specific studies. The fixed effects model was compared with a genome-wide random-effects model in which studies/disorders were allowed to have a different direction of effect.

Prediction of Allele-Specific Effects on Transcription

The overlap between polymorphic loci with microRNA binding sites was examined using the PolymiRTS 3.0 database (17). To check for the known influence of identified SNPs on gene expression, we searched a database of cis-acting eQTLs defined with RNA sequencing data of lymphoblastoid cell lines from 462 individuals, most of which were also examined in the 1000 Genomes Phase I dataset (18). Allele-specific transcription factor binding sites (TFBSs) were predicted with the web-based tool MatInspector version 2.1 (http://www.genomatix.de/online_help/help_matinspector/matinspector_help.html) (19). TFBS searches were performed for all promoter regions (40 kb upstream) of the top 100 SNPs identified in the full and the age-restricted meta-analyses (i.e., each allele flanked by 10 base pairs up- and downstream sequence). Sequences with a transcription factor–specific matrix core similarity of at least 0.75 were defined as potentially containing the respective TFBS.

RESULTS

SNP-Based Genetic Correlation

The SNP-based r_G between ADHD and BPD was substantial and significant for both the full and age-restricted samples. Interestingly, the r_G was higher for the age-restricted sample as compared with the full sample (r_{Gfull} = .64, SE = .02, $p = 3.13 \times 10^{-14}$; $r_{Grestricted}$ = .71, SE = .02, $p = 4.09 \times 10^{-16}$; Table 1).

Full Sample. Figure 1A shows the Manhattan plot of the primary cross-disorder meta-analysis, and Supplemental

Table 1. Results From the Univariate Heritability Analysis and Genetic Correlation Between Data From ADHD and BPD Complete and Age-Restricted Samples

BPD Cohort	*n* (Cases/Controls)	Heritability	SE	Genetic Correlation With ADHD	SE	*Z* Score	*p* Value
Age Restricted	5167/12,844	0.119	0.023	0.7062	0.087	7.593	4.09×10^{-16}
Full Sample	9650/12,844	0.068	0.024	0.6392	0.084	8.136	3.13×10^{-14}

Attention-deficit/hyperactivity disorder (ADHD): n_{cases} = 4609 and $n_{controls}$ = 8519; single nucleotide polymorphism–based h2 (observed) = 0.0652 (SE = 0.0279).

BPD, bipolar disorder; h2, heritability.

Figure S1A shows the corresponding quantile-quantile plot. Two independent loci, located on chromosomes 6 and 10, reached genome-wide significance ($p < 5 \times 10^{-8}$). The strongest signal ($p = 2.47 \times 10^{-8}$) was for SNP rs7089973, located on chromosome 10 in an intronic region of the TAF9B RNA polymerase II, TATA box binding protein–associated factor, 31 kDa pseudogene 2 gene (*TAF9BP2*; Figure 2A), which is a pseudogene of a yet unknown function. SNP rs7089973 is in LD with SNPs in the TruB pseudouridine synthase homolog 1 gene (*TRUB1*; best $p = 8.89 \times 10^{-6}$) and with the family with sequence similarity 160 member B1 gene (*FAM160B1*; best $p = 5.45 \times 10^{-8}$).

On chromosome 6, the strongest signal was for SNP rs11756438, located in an intronic region of the centrosomal protein 85 kDa–like gene (*CEP85L*; $p = 4.36 \times 10^{-8}$; Figure 2B) encoding a protein of unknown function. Furthermore, rs11756438 is in LD with SNPs in the bromodomain containing 7 pseudogene 3 gene (*BRD7P3*; best $p = 9.39 \times 10^{-7}$), the phospholamban gene (*PLN*; best $p = 7.25 \times 10^{-7}$), and the solute carrier family 35, member F1 gene (*SLC35F1*; best $p = 1.00 \times 10^{-5}$). Both associated regions showed the same direction of effect in ADHD and BPD. The disorder-specific contribution to each genome-wide significant locus can be found in Table 2, and forest plots from individual GWASs are shown in Supplemental Figure S5. Top-ranked SNPs for this analysis ($p < 10^{-6}$) are summarized in Supplemental Table S3.

Age-Restricted BPD Sample. The Manhattan plot for the age-restricted analysis is shown in Figure 1B; the respective

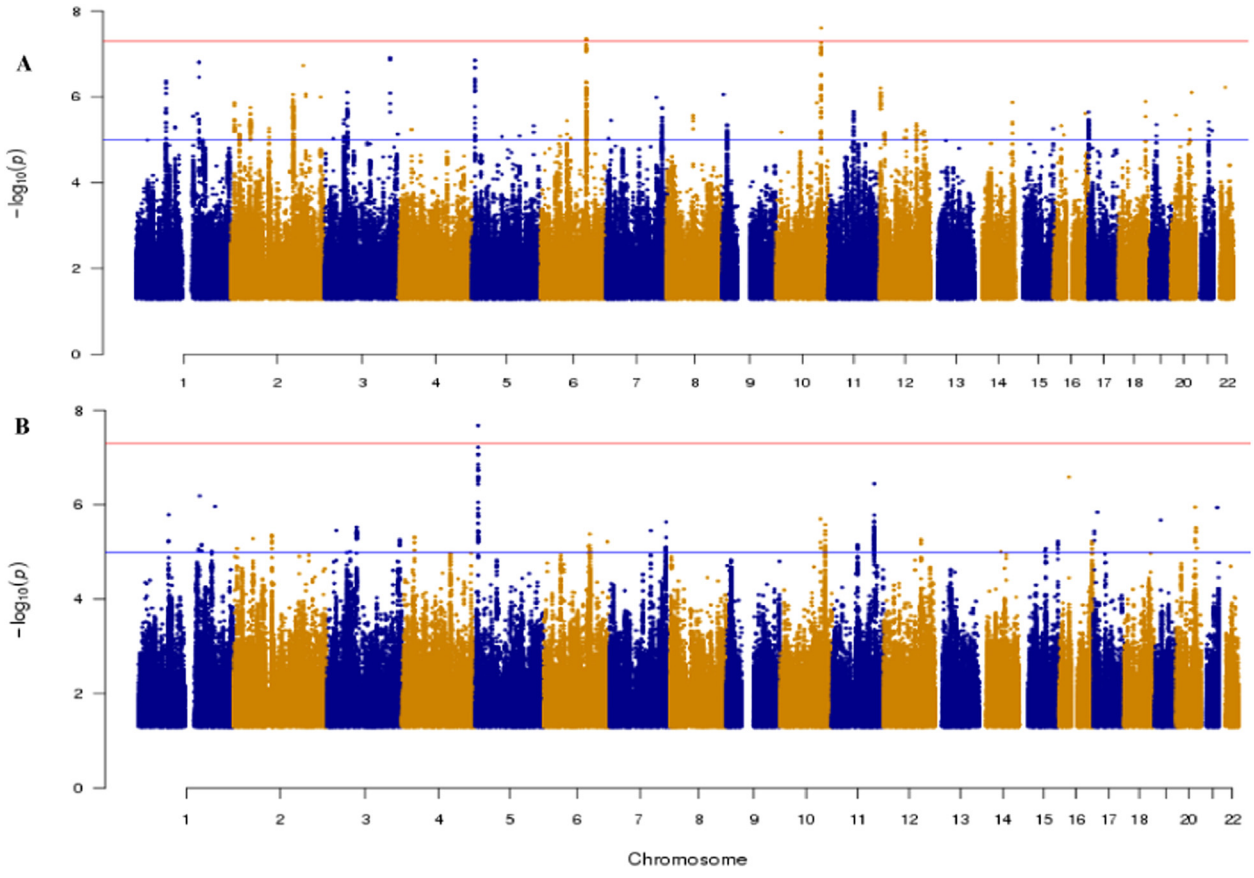

Figure 1. Manhattan plot of the **(A)** primary and **(B)** restricted cross-disorder meta-analyses. Only single nucleotide polymorphisms with *p* values ≤.05 are shown; horizontal lines show threshold for genome-wide significance ($p < 5 \times 10^{-8}$ in red) and suggestive association ($p < 1 \times 10^{-6}$ in blue).

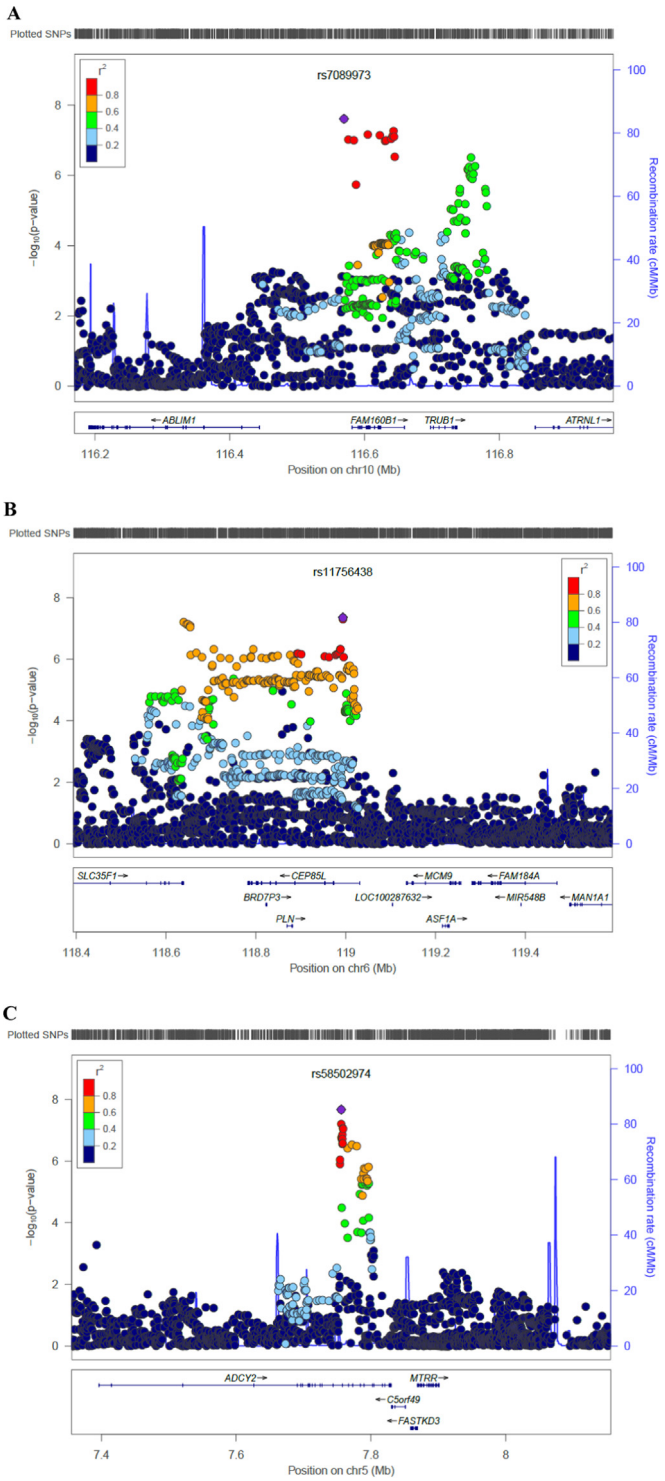

Figure 2. **(A)** Regional plot for genome-wide significant locus on chromosome (chr) 10 for the primary meta-analysis. **(B)** Regional plot for genome-wide significant locus on chr6 for the primary meta-analysis. **(C)** Regional plot for genome-wide significant locus on chr5 for the restricted meta-analysis. Results are shown as –log (*p* value) for genotyped and imputed single nucleotide polymorphisms (SNPs). The SNP showing strongest association is shown in the purple circle. The color of the remaining markers reflects r2 of the strongest associated SNP. The recombination rate is plotted in blue. cM/Mb, centimorgan/megabase.

quantile-quantile plot is provided in Supplemental Figure S1B. One SNP on chromosome 5 reached genome-wide significance; forest plots of results from individual GWAS are shown in Supplemental Figure S5. The strongest signal ($p = 2.47 \times 10^{-8}$) was observed for SNP rs58502974, located in an intron of the adenylate cyclase 2 (brain) (*ADCY2*) gene (Figure 2C). The product of this gene is a member of the family of adenylate cyclases, which are membrane-associated enzymes that catalyze the formation of the secondary messenger cyclic adenosine monophosphate (cAMP). Top-ranked SNPs

Table 2. Association Results Showing Effect Size and *p* for Genome-wide Significant Loci by Disorder for the Primary and Restricted Meta-analyses

SNP	Location (bp:chr)	A1/A2	MAF	OR_{ADHD}	p_{ADHD}	SE_{ADHD}	OR_{BPD}	p_{BPD}	SE_{BPD}	OR_{MA}	p_{MA}	Gene
Primary Meta-analysis												
rs7089973	10:116569565	A/C	0.36	0.90	1.56×10^{-3}	0.03	0.90	4.39×10^{-6}	0.02	0.90	2.47×10^{-8}	*TAF9BP2*
rs11756438	6:118993632	A/C	0.34	1.11	6.14×10^{-4}	0.03	1.11	1.85×10^{-5}	0.02	1.10	4.36×10^{-8}	*CEP85L*
Age-Restricted Meta-analysis												
rs58502974	5:7755900	A/T	0.44	1.14	1.84×10^{-5}	0.03	1.11	2.33×10^{-4}	0.03	1.13	2.11×10^{-8}	*ADCY2*

ADHD, attention-deficit/hyperactivity disorder; bp, base pair; BPD, bipolar disorder; chr, chromosome; MA, meta-analysis; MAF, minor allele frequency; OR, odds ratio; SNP, single nucleotide polymorphism.

for this analysis ($p < 10^{-4}$) are summarized in Supplemental Table S3.

Allele-Specific Transcriptional Activity

None of the top 100 associated SNPs in the primary and restricted analyses (Supplemental Table S3) were listed in the GEUVADIS (Genetic European Variation in Disease) database of cis-eQTLs defined in lymphoblastoid cell lines that were mainly derived from the 1000 Genomes sample (18). To estimate the potential eQTL function in other tissues, direct bioinformatics prediction was thus used and found differential binding of transcription factors for 13 markers in the primary and four in the restricted GWAS, which involved neurorelevant transcription factors at seven and one sites, respectively (Supplemental Table S5). A notable finding among those was for the gene *ABLIM1*. Allele-specific binding sites of microRNAs at the three prime untranslated region (3′-UTR) of family with sequence similarity 160, member B1 (*FAM160B1*) transcripts were also observed (Supplemental Table S5).

Extending this analysis to evaluate top-ranked SNPs ($p < 10^{-4}$) for effects on transcriptional activity, we queried the GEUVADIS database (18) for known eQTLs. Of the 4806 SNPs displaying a trend toward association ($p < 1 \times 10^{-4}$) in one of the two analyses, 192 indeed were known eQTLs (Supplemental Table S4). Of those, 74 markers influenced the expression of 5′-nucleotidase domain containing 2 (*NT5DC2*); the expression of its homolog *NT5DC1* was modulated by 17 SNPs. Two SNPs were found to influence the expression of the calcium-channel, voltage-dependent, beta 3 subunit (*CACNB3*). The accumulation of eQTLs at the *NT5DC* loci especially may be explained by LD. Indeed, when we tested the total number of eQTLs, it did not exceed expectation in either the full or the restricted analysis (odds $\text{ratio}_{\text{primary}} = 0.91$, $p_{\text{primary}} = 0.27$; odds $\text{ratio}_{\text{restricted}} = 0.97$, $p_{\text{restricted}} = 0.84$).

DISCUSSION

In this study, we set out to identify shared genetic risk factor for ADHD and BPD through cross-disorder meta-analysis of the existing GWAS samples from the Psychiatric Genomics Consortium. We hypothesized that overlap between the disorders might be most pronounced in BPD cases with an early age of onset, and that restricting the analysis to samples with an onset ≤21 years of age would increase the power of gene finding by reducing heterogeneity (8). Our findings clearly show the substantial and significant genetic correlation between ADHD and BPD, which supports the existence of genome-wide significant, shared genetic risk variants for ADHD and BPD ($r_{Gfull} = .64$, SE = .08, $p = 3.13 \times 10^{-14}$; $r_{Grestricted} = .71$, SE = .09, $p = 4.09 \times 10^{-16}$; Table 1). We contrasted these results by estimating the genetic correlation between ADHD and BPD using previously published data sets and found the same correlation pattern (Supplemental Table S2).

In the full sample, we found two genome-wide significant loci and 10 with suggestive evidence of association ($p < 10^{-6}$; Supplemental Table S3). Although the statistical power was potentially lower in the age-restricted sample, one significant finding was observed. The region identified was different from the top findings in the primary analysis, raising the possibility of development-specific gene activity in these two conditions. When specifically considering the BPD analyses (Supplemental Table S3; also compare Supplemental Figure S2A, B), it becomes clear that almost all top cross-disorder SNPs have lower *p* values in the age-restricted BPD sample as compared with the full BPD sample, despite its lower power. This might point to a stronger genetic component of early-onset BPD and consequently a different etiology as compared with late-onset BPD (20). However, these findings are still preliminary, as addressing this issue was not the major aim of our analysis, and further work is clearly needed to adequately answer such questions.

Our top hit in the full analysis was present on chromosome 10 in the gene *TAF9BP2*. This being a pseudogene of unknown function, we cannot speculate about its potential involvement in the ADHD-BPD covariation. *TRUB1*, which includes variants in LD with the top SNP, may constitute a potential candidate gene; its product is a member of the pseudouridine synthase gene family and may function as RNA chaperone, altering aspects of messenger RNA metabolism known to be affected by RNA structure (21). The protein encoded by *CEP85L*, suggested to be involved by the hit on chromosome 6, was identified as a breast cancer antigen (22); the gene was also associated in a meta-analysis with the myocardial repolarization (23,24), the latter lending some support to the notion that the gene's product is involved in neural conduction. This is the first time that genes in these loci are associated with psychiatric diseases, so the mechanisms by which variants in those might affect disease risk remain elusive. Our eQTL analyses did not provide evidence for a direct effect of the variation on the expression of the implicated genes or surrounding genes in LD with the hits. Furthermore, the functions of these genes are not well described, highlighting the need for further research.

The best hit found in the age-restricted sample is more obviously functionally significant. *ADCY2*, which codes for

adenylate cyclase 2, is a key regulator of cAMP metabolism and thus of second messaging by activating protein kinase A, thereby triggering cAMP response element binding protein phosphorylation (25). Both the protein kinase A and cAMP response element binding protein pathways (26,27) have been implicated in BPD, and *CREB1* is a well-documented candidate gene for BPD (28). Recently, an SNP in *ADCY2* was also found to be genome-wide significantly associated with BPD (29). There is a vast literature on disturbances of serotonin and dopamine signaling in ADHD; both monoamines target G protein–coupled receptors that use ADCY2 in their signal transduction cascades, so it is conceivable that this protein is also relevant to ADHD. As this association became significant ($p = 4.5 \times 10^{-8}$) when restricting the BPD sample to an age of onset ≤21 years old, the signal likely may be associated with a more neurodevelopmental form of BPD. One hypothesis for future research is that adenylate cyclase signaling is a core mechanism mediating the comorbidity between ADHD and BPD.

Our primary meta-analysis yielded 10 suggestive loci ($p < 10^{-6}$). In addition to the aforementioned genes, these loci comprise the following candidate genes, each supported by at least three suggestive associations: On chromosome 1, we observed a suggestive association with the retinoid X receptor, gamma gene (*RXRG*). *RXRG* codes for retinoid receptor gamma—a regulator of dopamine signaling, cocaine response, and affective behaviors in mice (30,31). This makes it an attractive candidate for both BPD and ADHD. The gene has been suggested to have an effect on hippocampal volume by QTL analysis in mice (32) and has been associated with sensation seeking (33), a personality trait associated with combined-type ADHD (34). On chromosome 3, a signal for the neuroligin 1 gene (*NLGN1*) was also found. The neuroligin protein family has already been implicated in a wide range of neuropsychiatric disorders including autism, schizophrenia, and BPD (35).

Presence of eQTLs was evaluated in SNPs with association p values $<10^{-4}$ in either of the cross-disorder GWAS meta-analyses (Supplemental Table S5). Several interesting candidate genes for ADHD-BPD covariation were found to be regulated by multiple SNPs in our list serving as eQTLs. Of greatest interest are the findings implicating two members of the NT5DC family, a family of haloacid dehalogenase–type phosphatases (36). *NT5DC1* has been associated with BPD in several studies, showing the strongest association with the disorder in National Institute of Mental Health Genetics Initiative bipolar pedigrees (37). Importantly, we recently observed an association between rare variants in this gene and adult ADHD (38). *NT5DC2* has previously been found associated with schizophrenia; furthermore, *NT5DC2* is a target of microRNA-137 and is differentially methylated as a function of childhood maltreatment in BPD patients (39). Another interesting finding was for *CACNB3*, which codes for a voltage-gated calcium channel involved in neuronal morphology and differentiation; its transcript is targeted by microRNA-34a, which is upregulated in cerebellum of bipolar patients (40). Allelic variation in *ABLIM1*, which has previously been associated with novelty seeking, harm avoidance, reward, and alcohol dependence (41), also appears to be a plausible finding.

Our finding that putative promoter regions containing the top-ranked associated SNPs do not contain known eQTLs was against our predictions of allele-specific effects on transcription. This discordance may, on the one hand, be attributed to the known high false positive rate of pattern searches with position-specific scoring matrices (19). On the other hand, the eQTL dataset was derived from analyses of lymphoblastoid cell lines (18), and it is therefore conceivable that the allele-specific TFBSs may serve as functional eQTLs in a different tissue context.

The findings described here need to be interpreted in the light of several caveats. The differential diagnosis especially between BPD and adult ADHD can be challenging. Although this does not affect the ADHD sample, which was ascertained in childhood, it might pose a problem for the BPD sample: some shared signals might be due to patients with adult ADHD having falsely received a diagnosis of BPD. Given that all studies included here relied on DSM inclusion criteria, we are confident that misdiagnosis is at least not common enough to account for genome-wide significant hits. Ethnic heterogeneity might influence the results as well, although we aimed to control for this by including multidimensional scaling components in every GWAS included in the meta-analyses. Finally, sample sizes in our cross-disorder analyses were still rather small for complex genetic traits. This was particularly damaging in the restricted analysis, where the reduction in sample size masked the potential increase in power due to the reduction in etiological heterogeneity (discussed above).

In conclusion, we provide evidence for the genetic overlap between ADHD and BPD. This genome-wide SNP-based genetic overlap confirms the involvement of pathways such as G protein–coupled signaling already known for their role in hyperactivity and/or emotional behaviors, and implicate a new candidate pathway (i.e., messenger RNA stability) in the pathophysiology of both ADHD and BPD.

ACKNOWLEDGMENTS AND DISCLOSURES

This work was supported by core funding for the Psychiatric Genomics Consortium provided by the National Institute of Mental Health (NIMH) (Grant No. U01 MH094421). This work was also supported by grants from the NIMH (Grant Nos. MH081804, MH078151, and MH59567 to JRK).

This work was carried out on the Dutch national e-infrastructure with the support of the SURF Foundation. We especially thank Willem Vermin (SURF Foundation, Amsterdam, the Netherlands) for his valuable support in the creating computer codes that fulfilled our needs for an efficient analysis of the data.

SVF is supported by the K.G. Jebsen Centre for Research on Neuropsychiatric Disorders, University of Bergen, Bergen, Norway, the European Community's Seventh Framework Programme (FP7/2007-2013) under Grant No. 602805 and NIMH Grant Nos. R13MH059126 and R01MH094469. JRK is supported by National Institutes of Health Grant Nos. MH078151, MH081804, MH59567, and MH094483. AR is supported by the Deutsche Forschungsgemeinschaft (Grant Nos. KFO 125, TRR 58/B06 and Z02, RE1632/5-1, and RTG 1256) and the European Community's Seventh Framework Programme (FP7/2007-2013) under Grant No. 602805 ("AGGRESSOTYPE"). CJS and HW are supported by Interdisziplinäres Zentrum für Klinische Forschung (IZKF) Grant No. Z-6. KJEvH, BF, and AA-V are supported by grants from the Netherlands Organization for Scientific Research (NWO), i.e., the NWO Brain & Cognition Excellence Program (Grant No. 433-09-229) and a Vici grant to BF (Grant No. 016-130-669), and by grants from the Netherlands Brain Foundation (Grant No. 15F07[2]27) and BBMRI-NL (Grant No. CP2010-33). The research leading to these results also received funding from the European Community's

Seventh Framework Programme (FP7/2007-2013) under Grant Nos. 602805 (AGGRESSOTYPE), 278948 (TACTICS), and 602450 (IMAGEMEND), and from the European Community's Horizon 2020 Programme (H2020/2014-2020) under Grant Nos. 643051 (MiND) and 667302 (CoCA). In addition, their work is supported by the ECNP for the Research Network "ADHD across the Lifespan."

BF received an educational speaking fee from Merz. EJS-B received fees for speaking, consultancy, research funding and conference support from Shire Pharma, speaker fees from Janssen Cilag, Medice, book royalties from OUP and Jessica Kingsley and consultancy from Neurotech solutions. ML declares that, over the past 36 months, he has received lecture honoraria from Biophausia Sweden, Servier Sweden, AstraZeneca, and served at advisory board for Lundbeck Pharmaceuticals. No other equity ownership, profit-sharing agreements, royalties, or patent. OAA received speakers' honoraria from Lundbeck and GSK. In the past year, SVF received income, travel expenses, potential income and/or research support from Pfizer, Ironshore, Shire, Akili Interactive Labs, CogCubed, Alcobra, VAYA Pharma, Neurovance, Impax, NeuroLifeSciences. With his institution, he has US patent US20130217707 A1 for the use of sodium-hydrogen exchange inhibitors in the treatment of ADHD. He receives royalties from books published by Guilford Press: *Straight Talk about Your Child's Mental Health*, Oxford University Press: *Schizophrenia: The Facts* and Elsevier, *ADHD: Non-Pharmacologic Interventions*. All other authors report no biomedical financial interests or potential conflicts of interest.

Composition of the Psychiatric Genomics Working Groups - ADHD Working Group

Richard J.L. Anney, Alejandro Arias Vasquez, Maria J. Arranz, Philip Asherson, Tobias J. Banaschewski, Mònica Bayés, Joseph Biederman, Jan K. Buitelaar, Miguel Casas, Alice Charach, Bru Cormand, Jennifer Crosbie, Soeren Dalsgaard, Mark J. Daly, Alysa E. Doyle, Richard P. Ebstein, Josephine Elia, Stephen V. Faraone, Barbara Franke, Christine Freitag, Michael Gill, Hakon Hakonarson, Amaia Hervas, Peter Holmans, Lindsey Kent, Jonna Kuntsi, Nanda Lambregts-Rommelse, Kate Langley, Klaus-Peter Lesch, Sandra K. Loo, Joanna Martin, James J. McGough, Sarah E. Medland, Jobst Meyer, Eric Mick, Ana Miranda, Fernando Mulas, Benjamin M. Neale, Stan F. Nelson, Michael C. O'Donovan, Robert D. Oades, Michael J. Owen, Haukur Palmason, Josep A. Ramos-Quiroga, Andreas Reif, Marta Ribasés, Stephan Ripke, Herbert Roeyers, Jasmin Romanos, Marcel Romanos, Aribert Rothenberger, Cristina Sánchez-Mora, Russell Schachar, Joseph Sergeant, Susan L. Smalley, Edmund J.S. Sonuga-Barke, Hans-Christoph Steinhausen, Anita Thapar, Alexandre Todorov, Susanne Walitza, Yufeng Wang, Andreas Warnke, Nigel Williams, Yanli Zhang-James

Bipolar Disorder Working Group

Devin Absher, Huda Akil, Ole A. Andreassen, Adebayo Anjorin, Lena Backlund, Judith A. Badner, Jack D. Barchas, Thomas B. Barrett, Nick Bass, Michael Bauer, Frank Bellivier, Sarah E. Bergen, Wade Berrettini, Douglas Blackwood, Cinnamon S. Bloss, Michael Boehnke, Gerome Breen, René Breuer, William E. Bunney, Margit Burmeister, William Byerley, Sian Caesar, Kim Chambert, Sven Cichon, David A Collier, Aiden Corvin, William Coryell, Nick Craddock, David W. Craig, Mark Daly, Richard Day, Franziska Degenhardt, Srdjan Djurovic, Frank Dudbridge, Howard J. Edenberg, Amanda Elkin, Bruno Etain, Anne Farmer, Manuel Ferreira, I. Nicol Ferrier, Matthew Flickinger, Tatiana Foroud, Josef Frank, Christine Fraser, Louise Frisén, Janice Fullerton, Elliot S. Gershon, Michael Gill, Katherine Gordon-Smith, Elaine K. Green, Tiffany A. Greenwood, Detelina Grozeva, Weihua Guan, Hugh Gurling, Ómar Gustafsson, Marian L. Hamshere, Martin Hautzinger, Chantal Henry, Stefan Herms, Maria Hipolito, Peter A. Holmans, Christina Hultman, Stéphane Jamain, Edward G. Jones, Ian R. Jones, Lisa Jones, Jean-Pierre Kahn, Radhika Kandaswamy, John R. Kelsoe, James L. Kennedy, George Kirov, Daniel L. Koller, Phoenix Kwan, Mikael Landén, Niklas Langstrom, Mark Lathrop, Jacob Lawrence, William B. Lawson, Marion Leboyer, Phil H. Lee, Jun Li, Paul Lichtenstein, Danyu Lin, Chunyu Liu, Falk W. Lohoff, Susanne Lucae, Pamela B. Mahon, Sandra Maier, Wolfgang Maier, Nick Martin, Manuel Mattheisen, Keith Matthews, Morten Mattingsdal, Kevin McGhee, Peter McGuffin, Melvin G. McInnis, Andrew McIntosh, Rebecca McKinney, Alan W. McLean, Francis J. McMahon, Andrew McQuillin, Ingrid Melle, Fan Guo Meng, Philip B. Mitchell, Grant W. Montgomery, Jennifer Moran, Gunnar Morken, Derek Morris, Valentina Moskvina, Pierandrea Muglia, Thomas W. Mühleisen, Walter J. Muir, Bertram Müller-Mysok, Richard M. Myers, Caroline M. Nievergelt, Ivan Nikolov, Vishwajit Nimgaonkar, Markus M. Nöthen, John I. Nurnberger, Jr., Evaristus A. Nwulia, Michael C. O'Donovan, Colm O'Dushlaine, Urban Osby, Högni Óskarsson, Michael J. Owen, Roy Perlis, Hannes Petursson, Benjamin S. Pickard, James B. Potash, Peter Propping, Shaun Purcell, Emma Quinn, Soumya Raychaudhuri, Andreas Reif, John Rice, Marcella Rietschel, Stephan Ripke, Douglas Ruderfer, Martin Schalling, Alan F. Schatzberg, William A. Scheftner, Peter R. Schofield, Nicholas J. Schork, Thomas G. Schulze, Johannes Schumacher, Markus Schwarz, Ed Scolnick, Laura J. Scott, Paul D. Shilling, Engilbert Sigurðsson, Pamela Sklar, Erin N. Smith, Jordan Smoller, David St. Clair, Hreinn Stefansson, Kari Stefansson, Michael Steffens, Stacy Steinberg, John Strauss, Jana Strohmaier, Patrick Sullivan, Szabolcs Szelinger, Robert C. Thompson, Þorgeir Þorgeirsson, Federica Tozzi, Jens Treutlein, John B. Vincent, Stanley J. Watson, Thomas F. Wienker, Richard Williamson, Stephanie H. Witt, Adam Wright, Wei Xu, Allan H Young, Peter P. Zandi, Peng Zhang, Sebastian Zöllner

See the Supplement for affiliations of all authors in the PGC working groups.

ARTICLE INFORMATION

From the Department of Human Genetics (KJEvH, BF, MK, AA-V), Department of Psychiatry (BF, AA-V), and Department of Cognitive Neuroscience (AA-V), Radboud University Medical Center; and Donders Institute for Brain, Cognition and Behaviour (KJEvH, MK, AA-V), Radboud University, Nijmegen, the Netherlands; Core Unit Systems Medicine (CJS), University of Würzburg; Department of Psychiatry (K-PL, HW), Psychosomatics and Psychotherapy, University Hospital of Würzburg, Würzburg; and Department of Psychiatry, Psychosomatic Medicine and Psychotherapy (HW, AR), University Hospital of Frankfurt, Frankfurt, Germany; Analytic and Translational Genetics Unit (SR), Massachusetts General Hospital, Boston, Massachusetts; Division of Psychiatry (AQ), University College London, London; and Department of Psychology (EJS-B), University of Southampton, Southampton, United Kingdom; Department of Psychiatry (JRK), University of California, San Diego, San Diego, California; The Institute of Neuroscience and Physiology (ML), University of Gothenburg, Gothenburg; and Department of Medical Epidemiology and Biostatistics (ML), Karolinska Institutet, Stockholm, Sweden; NORMENT – K.G. Jebsen Centre for Psychosis Research (OAA), Division of Mental Health and Addiction, Oslo University Hospital & Institute of Clinical Medicine, University of Oslo, Oslo; and K.G. Jebsen Centre for Neuropsychiatric Disorders (SVF), Department of Biomedicine, University of Bergen, Bergen, Norway; and the Departments of Psychiatry and Neuroscience and Physiology (SVF), SUNY Upstate Medical University, Syracuse, New York.

KJEvH and CJS contributed equally to this work.

AA-V and AR shared final responsibility for this work.

Address correspondence to Alejandro Arias-Vásquez, MSc., DSc., Ph.D., Radboudumc, Department of Human Genetics, route 855, Postbus 9101, 6500 HB Nijmegen, the Netherlands; E-mail: alejandro.ariasvasquez@radboudumc.nl.

Received Apr 8, 2016; revised July 11, 2016; accepted Aug 8, 2016.

Supplementary material cited in this article is available online at http://dx.doi.org/10.1016/j.biopsych.2016.08.040.

REFERENCES

1. Faraone SV, Perlis RH, Doyle AE, Smoller JW, Goralnick JJ, Holmgren MA, *et al*. (2005): Molecular genetics of attention-deficit/hyperactivity disorder. Biol Psychiatry 57:1313–1323.
2. Shih RA, Belmonte PL, Zandi PP (2004): A review of the evidence from family, twin and adoption studies for a genetic contribution to adult psychiatric disorders. Int Rev Psychiatry 16:260–283.
3. Identification of risk loci with shared effects on five major psychiatric disorders (2013): A genome-wide analysis. Lancet 381:1371–1379.
4. Klassen LJ, Katzman MA, Chokka P (2010): Adult ADHD and its comorbidities, with a focus on bipolar disorder. J Affect Disord 124: 1–8.
5. Gross-Lesch S, Dempfle A, Reichert S, Jans T, Geissler J, Kittel-Schneider S, *et al*. (2016): Sex- and subtype-related differences in the comorbidity of adult ADHDs. J Atten Disord 20:855–866.

6. Faraone SV, Asherson P, Banaschewski T, Biederman J, Buitelaar JK, Ramos-Quiroga JA, *et al*. (2015): Attention-deficit/hyperactivity disorder. Nat Rev Dis Primers 1:15020.
7. Wingo AP, Ghaemi SN (2007): A systematic review of rates and diagnostic validity of comorbid adult attention-deficit/hyperactivity disorder and bipolar disorder. J Clin Psychiatry 68:1776–1784.
8. Ryden E, Thase ME, Straht D, Aberg-Wistedt A, Bejerot S, Landen M (2009): A history of childhood attention-deficit hyperactivity disorder (ADHD) impacts clinical outcome in adult bipolar patients regardless of current ADHD. Acta Psychiatr Scand 120:239–246.
9. Lee SH, Ripke S, Neale BM, Faraone SV, Purcell SM, Perlis RH, *et al*. (2013): Genetic relationship between five psychiatric disorders estimated from genome-wide SNPs. Nat Genet 45:984–994.
10. Schimmelmann BG, Hinney A, Scherag A, Putter C, Pechlivanis S, Cichon S, *et al*. (2013): Bipolar disorder risk alleles in children with ADHD. J Neural Transm 120:1611–1617.
11. Landaas ET, Johansson S, Halmoy A, Oedegaard KJ, Fasmer OB, Haavik J (2011): Bipolar disorder risk alleles in adult ADHD patients. Genes Brain Behav 10:418–423.
12. Manchia M, Adli M, Akula N, Ardau R, Aubry JM, Backlund L, *et al*. (2013): Assessment of response to lithium maintenance treatment in bipolar disorder: A Consortium on Lithium Genetics (ConLiGen) report. PLoS One 8:e65636.
13. Neale BM, Medland S, Ripke S, Anney RJ, Asherson P, Buitelaar J, *et al*. (2010): Case-control genome-wide association study of attention-deficit/hyperactivity disorder. J Am Acad Child Adolesc Psychiatry 49:906–920.
14. Sklar P, Ripke S, Scott LJ, Andreassen OA, Cichon S, Craddock N, *et al*. (2011): Large-scale genome-wide association analysis of bipolar disorder identifies a new susceptibility locus near ODZ4. Nat Genet 43:977–983.
15. Bulik-Sullivan BK, Loh PR, Finucane HK, Ripke S, Yang J, Schizophrenia Working Group of the Psychiatric Genomics Consortium, *et al*. (2015): LD score regression distinguishes confounding from polygenicity in genome-wide association studies. Nat Genet 47:291–295.
16. Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MA, Bender D, *et al*. (2007): PLINK: A tool set for whole-genome association and population-based linkage analyses. Am J Hum Genet 81:559–575.
17. Bhattacharya A, Ziebarth JD, Cui Y (2014): PolymiRTS Database 3.0: Linking polymorphisms in microRNAs and their target sites with human diseases and biological pathways. Nucleic Acids Res 42:D86–D91.
18. Lappalainen T, Sammeth M, Friedlander MR, t Hoen PA, Monlong J, Rivas MA, *et al*. (2013): Transcriptome and genome sequencing uncovers functional variation in humans. Nature 501:506–511.
19. Cartharius K, Frech K, Grote K, Klocke B, Haltmeier M, Klingenhoff A, *et al*. (2005): MatInspector and beyond: promoter analysis based on transcription factor binding sites. Bioinformatics 21:2933–2942.
20. Faraone SV, Glatt SJ, Tsuang MT (2003): The genetics of pediatric-onset bipolar disorder. Biol Psychiatry 53:970–977.
21. Carlile TM, Rojas-Duran MF, Zinshteyn B, Shin H, Bartoli KM, Gilbert WV (2014): Pseudouridine profiling reveals regulated mRNA pseudouridylation in yeast and human cells. Nature 515:143–146.
22. Wu X, Scelo G, Purdue MP, Rothman N, Johansson M, Ye Y, *et al*. (2012): A genome-wide association study identifies a novel susceptibility locus for renal cell carcinoma on 12p11.23. Hum Mol Genet 21:456–462.
23. Ritchie MD, Denny JC, Zuvich RL, Crawford DC, Schildcrout JS, Bastarache L, *et al*. (2013): Genome- and phenome-wide analyses of cardiac conduction identifies markers of arrhythmia risk. Circulation 127:1377–1385.
24. Newton-Cheh C, Eijgelsheim M, Rice KM, de Bakker PI, Yin X, Estrada K, *et al*. (2009): Common variants at ten loci influence QT interval duration in the QTGEN Study. Nat Genet 41:399–406.
25. Delghandi MP, Johannessen M, Moens U (2005): The cAMP signalling pathway activates CREB through PKA, p38 and MSK1 in NIH 3T3 cells. Cell Signal 17:1343–1351.
26. Nurnberger JI Jr., Koller DL, Jung J, Edenberg HJ, Foroud T, Guella I, *et al*. (2014): Identification of pathways for bipolar disorder: A meta-analysis. JAMA Psychiatry 71:657–664.
27. Kerner B, Rao AR, Christensen B, Dandekar S, Yourshaw M, Nelson SF (2013): Rare genomic variants link bipolar disorder with anxiety disorders to CREB-regulated intracellular signaling pathways. Front Psychiatry 4:154.
28. Li M, Luo XJ, Rietschel M, Lewis CM, Mattheisen M, Muller-Myhsok B, *et al*. (2014): Allelic differences between Europeans and Chinese for CREB1 SNPs and their implications in gene expression regulation, hippocampal structure and function, and bipolar disorder susceptibility. Mol Psychiatry 19:452–461.
29. Muhleisen TW, Leber M, Schulze TG, Strohmaier J, Degenhardt F, Treutlein J, *et al*. (2014): Genome-wide association study reveals two new risk loci for bipolar disorder. Nat Commun 5:3339.
30. Krzyzosiak A, Szyszka-Niagolov M, Wietrzych M, Gobaille S, Muramatsu S, Krezel W (2010): Retinoid x receptor gamma control of affective behaviors involves dopaminergic signaling in mice. Neuron 66:908–920.
31. Krezel W, Ghyselinck N, Samad TA, Dupe V, Kastner P, Borrelli E, *et al*. (1998): Impaired locomotion and dopamine signaling in retinoid receptor mutant mice. Science 279:863–867.
32. Lu L, Airey DC, Williams RW (2001): Complex trait analysis of the hippocampus: Mapping and biometric analysis of two novel gene loci with specific effects on hippocampal structure in mice. J Neurosci 21:3503–3514.
33. Alliey-Rodriguez N, Zhang D, Badner JA, Lahey BB, Zhang X, Dinwiddie S, *et al*. (2011): Genome-wide association study of personality traits in bipolar patients. Psychiatr Genet 21:190–194.
34. Jacob CP, Gross-Lesch S, Reichert S, Geissler J, Jans T, Kittel-Schneider S, *et al*. (2014): Sex- and subtype-related differences of personality disorders (Axis II) and personality traits in persistent ADHD. J Atten Disord 20:1056–1065.
35. Carroll LS, Owen MJ (2009): Genetic overlap between autism, schizophrenia and bipolar disorder. Genome Med 1:102.
36. Seifried A, Schultz J, Gohla A (2013): Human HAD phosphatases: Structure, mechanism, and roles in health and disease. FEBS J 280:549–571.
37. Bigdeli TB, Maher BS, Zhao Z, Sun J, Medeiros H, Akula N, *et al*. (2013): Association study of 83 candidate genes for bipolar disorder in chromosome 6q selected using an evidence-based prioritization algorithm. Am J Med Genet B Neuropsychiatr Genet 162B:898–906.
38. Zayats T, Jacobsen KK, Kleppe R, Jacob CP, Kittel-Schneider S, Ribasés M, *et al*. (2016): Exome chip analyses in adult attention deficit hyperactivity disorder. Transl Psychiatry 6:e923.
39. Prados J, Stenz L, Courtet P, Prada P, Nicastro R, Adouan W, *et al*. (2015): Borderline personality disorder and childhood maltreatment: A genome-wide methylation analysis. Genes Brain Behav 14:177–188.
40. Bavamian S, Mellios N, Lalonde J, Fass DM, Wang J, Sheridan SD, *et al*. (2015): Dysregulation of miR-34a links neuronal development to genetic risk factors for bipolar disorder. Mol Psychiatry 20:573–584.
41. Wang KS, Liu X, Aragam N, Mullersman JE, Jian X, Pan Y, *et al*. (2012): Polymorphisms in ABLIM1 are associated with personality traits and alcohol dependence. J Mol Neurosci 46:265–271.